CLAIMS

1. A method for preparing an immunoreagent which goes into making up a calibration system applied to the assaying of multiple analytes in the same biological sample, said method using various categories of particles, each category of particles being sensitized by association with a ligand specific for one of the analytes to be assayed, said method characterized in that it comprises the steps which consist in:

a) determining, for each category of sensitized particles and for each of n given amounts of ligand associated with said particles, the curve of response as a function of the concentration of homologous compound, over a range of concentrations corresponding to the known measurement range of the analyte to be assayed;

b) selecting, for each category of sensitized particles, the curve corresponding to the smallest amount of ligand which gives a significant response signal and which is compatible with the use of a sample dilution and of a labeling reagent, which are common to all the analytes simultaneously assayed;

c) evaluating, for each category of particles sensitized, according to the curve selected in step b), the mean signal corresponding to the signal associated with a point characteristic of each of said curves, thus obtaining as many mean signals as there are categories of particles;

d) adjusting, where appropriate, the amounts of ligand associated with each category of particles such that all the mean signals evaluated in step c) are within a ratio of 1 to 5, and

e) mixing, in an appropriate solvent, the various categories of sensitized particles which correspond to the criterion of step d).

2. The method as claimed in claim 1, characterized in that, in step a), the amounts of ligand used vary by steps of 2 to 4.

3. The method as claimed in claim 1 or 2, characterized in that the sensitization of said particles with said ligands is carried out by covalence, by means of a biologically and/or chemically reactive intermediate molecular layer, or by using an affinity-based interaction system.

4. The method as claimed in any one of the preceding claims, characterized in that the concentration of homologous compound is expressed in biological units over a range which is identical for all the analytes.

5. The method as claimed in any one of the preceding claims, characterized in that the solvent used in step e) is suitable for and common to all the ligands.

6. The method as claimed in any one of the preceding claims, characterized in that said ligands consist of antigens and/or of antibodies.

7. An immunoreagent intended for the assaying of multiple analytes in biological samples, said reagent comprising a solvent and, mixed in with said solvent, various categories of particles, each of which is sensitized by association with a given amount of a ligand specific for one of the analytes to be assayed, characterized in that, for each of the categories of particles, and for a given concentration of compound homologous to the ligand, as expressed in biological units, said given amount of ligand results in a signal referred to as ``mean signal'', which is within a ratio of 1 to 5 with the mean signals obtained for the other categories of particles.

8. A kit for assaying multiple analytes in biological samples using various categories of particles, each category of particles being sensitized with a ligand specific for one of the analytes to be assayed, characterized in that it comprises:

i) an immunoreagent derived from the method as claimed in any one of claims 1 to 6,

ii) at least one calibration standard consisting of

a single homologous compound which reacts with one of the categories of particles which goes into making up the immunoreagent,

iii) a table of correspondence between the concentration, expressed in biological units, of the homologous compound constituting the calibration standard and that of each of the compounds homologous to the other ligands attached to the other categories of particles which go into making up the immunoreagent, and

iv) a labeling reagent.

9. The kit as claimed in claim 8, characterized in that said calibration standard comprises a homologous compound which reacts directly or indirectly with the ligand attached to the sensitized particle.

10. The kit as claimed in claim 8 or 9, characterized in that said homologous compound is of the same origin as the analyte to be assayed.

11. The kit as claimed in claim 8 or 9, characterized in that said homologous compound and the analyte to be assayed are of different origins.

12. The kit as claimed in any one of claims 8 to 11, characterized in that said labeling reagent consists of an immunocompound capable of quantifying the reaction between the analytes and the immunoreagent, said immunocompound being coupled to a label which is preferably a fluorochrome.

13. The kit as claimed in claim 12, characterized in that said labeling reagent reacts with the homologous compound or analyte to be assayed/ligand complex, in which case the assaying is direct, or with the uncomplexed ligand, in which case the assaying is indirect.

14. A method for using the kit as claimed in any one of claims 8 to 13, characterized in that it consists in measuring the signals resulting from the interaction between the immunoreagent and, firstly, the biological sample, and secondly, the calibration standard, and in determining and applying, to the various resulting signals,

a correction factor so as to obtain the titration, expressed in biological units, of each analyte of the sample, said correction factor being the ratio between the signal obtained for the calibration standard and the concentrations derived from the correspondence table.

15. The method as claimed in claim 14, characterized in that comprises the steps which consist in:

a) incubating, firstly, the biological sample and, secondly, the calibration standard with a predetermined amount of immunoreagent;

b) adding the labeling reagent;

c) measuring, by flow cytometry, the signals emitted, firstly, by the calibration standard and secondly, by the sample;

d) determining, for each category of particles sensitized, a correction factor which corresponds to the ratio between the concentration in biological units of each analyte to be assayed as given by the correspondence table and the signal measured in step c) for the calibration standard;

e) multiplying the signal emitted by each category of particles and measured in step c) by said correction factor, calculated for each analyte, so as to deduce therefrom the concentration in biological units of each analyte in the sample tested.

16. The method as claimed in claim 15, applied to the simultaneous and direct quantification of antibodies having different antigenic specificities, characterized in that said analytes to be assayed consist of antibodies and said ligands consist of antigens, and in that said labeling reagent consists of one or more second antibodies labeled with a fluorochrome which react specifically with the antibodies intended to be assayed.

17. The method as claimed in either of claims 15 and 16, applied to the simultaneous and direct quantification of various antigens, characterized in that said analytes to be assayed consist of antigens and said ligands consist of

antibodies, and in that said labeling reagent consists of a mixture of second antibodies labeled with a fluorochrome which react specifically with the antigens which are intended to be assayed.

18. The method as claimed in claim 15 or 16, applied to the simultaneous and indirect quantification of various antigens, characterized in that said analytes to be assayed consist of antigens and said ligands consist of antibodies, and in that said labeling reagent consists of a mixture of antigens labeled with a fluorochrome which compete with the analytes to be assayed for forming complexes with the ligands.

19. The method as claimed in claim 15 or 16, applied to the simultaneous and indirect quantification of various antibodies, characterized in that said analytes to be assayed consist of antibodies and said ligands consist of antigens, and in that said labeling reagent consists of a mixture of antibodies labeled with a fluorochrome which compete with the analytes to be assayed for forming complexes with the ligands.